IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s): Csöregi et al

Application No.: 10/019,651

Filed: 7/8/2002

Title: BIOSENSOR

Attorney Docket No.: 50159-026

Group Art Unit: 1651

Examiner: Jon P. Weber

Confirmation No:

DECLARATION UNDER 37 CFR 5 1.132

The undersigned hereby declares as follows:

- 1. We are named inventors of the above-referenced application, and we are familiar with the application, including the claims thereof.
- 2. We understand that an Official Action has issued in this case in which the Examiner takes the position that the enzyme from grass pea is more selective and sensitive than the enzyme from Aspergillus. We further understand that the method of the present invention would be relevant if this method results in a different, improved result compared to said obtained using Aspergillus.
- 3. Tests have been conducted to demonstrate the effects obtained using amine oxidase from grass pea, and from Aspergillus, respectively, particularly in demonstrating presence of histamine using transducers.

The following comparison data are present, where amine oxidase from grass pea have been compared with amine oxidase from Aspergillus niger. The comparison happens to be related to different transducer systems, but still it is apparent that amine oxidase from Aspergillus niger provides a lower sensitivity than amine oxidase from grass pea (0.93 vs. 0.48), as well as the same high potential is not needed (+200 mV vs. +600 mV). High potentials will

lead to high background currents and a small selectivity due to disturbing signals related to electro chemical interferences. A system comprising HRP (horse radish peroxidase) will become quite different, but provides on the other hand for a high selectivity with regard to putrescine and cadaverine. Amine oxidase from *Aspergillus niger* can not be used for putrescine, but then a peroxidase from *Micrococcus rubens* is needed.

ENZYME	TRANSDUCER	E vs. Ag/AgCl (mV)	SELECTIVITY	LR (µM)	DL (μM)	S (mA/M)
AO	Graphite	-50	His 100 %	1-150	0.33	5.16
grass pes			Put 147 %	1-400	0.17	13.58
HRP			Cad 132 %	1-400	0.20	11.80
AO	Graphite	+200	His 100 %	10-200	2.2	0.48
grass pea			Put < 1 %	-		
	·		Cad < 1 %		-	-
AO	I't paste	+600	His	0.17-20	ท.ล.	0.93
A. niger			Pur	0.06 200	n.a.	2.4

LR stands for Linear range

DL stands for Detection limit

S stands for Sensitivity

We hereby declare that all statements made herein of our own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

dated: 26/11 - 2003 dishaela Nusulascu

dated: 17/12 - 2003 dishaela Nusulascu

Ivo Frébort

Page 2